Remarks

Claims 1-38 are pending. Claims 5 and 30 have been amended. Claims 36-38 have been added. Claim 5 has been amended to more clearly recite what applicants consider to be their invention by deleting the phrase "on a chromosome of the cell". The amendment to claim 5 removes an unnecessary limitation and makes the language of claim 5 parallel with the language of claims 13 and 17. The amendment to claim 30 adopts a suggestion in the Office Action by requiring both an essential gene and a lethal gene. It is believed that this amendment causes, as suggested in the Office Action, claims 30-35 to read on the elected invention. It is believed that claims 31 and 32 read on species G and claims 30 and 33-35 are generic (however, see discussion below).

Claims 36-38 have been newly added. Each recites a cell (with an Environmentally Limited Viability System) in which the absence of a functional expression product of the regulatory gene derepresses expression of the lethal (claim 36), essential (claim 37), or replication (claim 38) gene, and is not expressed or inactive only in the non-permissive (claim 36) or permissive (claims 37, 38) environment. Support for claims 36-38 can be found at least on page 6, lines 21-25. Applicants submit that claims 36-38 represent the inverse of the mode of regulation recited in claims 6, 14, and 18, respectively (i.e. a lack of repression constitutes derepression). Accordingly, and pursuant to MPEP § 809.02(a), applicants assert that claims 36-38 correspond to species A, C, and E, respectively. A copy

of all of the pending claims, as they are believed to have been amended, are attached to this Amendment as an appendix.

Applicants also note that, with respect to species A and B, claims 1-5, 8-35, 37, and 38 are generic, with respect to species C and D, claims 1-13, 16-36, and 38 are generic, and with respect to species E and F, claims 1-17 and 20-37 are generic (see discussion below). With respect to species G, applicants note that claims 1-20, 23, 24, 27-30, and 33-38 are generic (see discussion below).

Response to Restriction Requirement

In response to the changed restriction requirement set forth in the Office Action mailed October 18, 1996, applicants elect for prosecution Group I, claims 1-29, with traverse. It is believed that in view of suggestions made in the Office Action and the amendments to the claims, all of the pending claims, claims 1-38, correspond to the elected invention. Applicants also provisionally elect species C, claims 13 and 14, with traverse, for prosecution. It is noted that newly added claim 37 reads on the elected species (see also discussion below).

In the Office Action mailed October 18, 1996, the claims were divided into 2 groups, Group I, claims 1-29, drawn to an isolated microbial cell, and Group II, claims 30-35, drawn to a method of inducing immunoprotection in an animal. The Office Action suggested that requiring both an essential gene and a lethal gene in the method of claim 30 would obviate the restriction requirement between the two groups. In response, applicants have amended

claim 30 to require both an essential gene and a lethal gene. Accordingly, applicants assert that all of the pending claims, claims 1-38, read on the elected invention. Applicants reserve the right to pursue claims to the subject matter of Group II in a separate application.

The Office Action also required election of a single species from among seven. These were Species A, claims 5 and 6, drawn to a cell having a regulatory gene down regulating the lethal gene in a permissive environment; Species B, claims 5 and 7, drawn to a cell having a regulatory gene up regulating the lethal gene in a non-permissive environment; Species C, claims 13 and 14, drawn to a cell having a regulatory gene down regulating the essential gene in a non-permissive environment; Species D, claims 13 and 15, drawn to a cell having a regulatory gene up regulating the essential gene in a permissive environment; Species E, claims 17 and 18, drawn to a cell having a regulatory gene down regulating the replication gene in a non-permissive environment; Species F, claims 17 and 19, drawn to a cell having a regulatory gene up regulating the replication gene in a permissive environment; and Species G, claims 21, 22, 25, and 26, drawn to a cell having an antigen expression gene.

Applicants initially note that the requirement for election of species appears to be improperly drawn in several respects. First, the seven species are not embodiments reciting mutually exclusive characteristics (see MPEP § 806.04(f)). For example, a cell of the invention can have, independently, a lethal gene, an essential gene, and a replication gene, each under independent regulation (in fact, claim 1 requires the presence of both an essential

gene and a lethal gene). Furthermore, any cell having any of the regulatory schemes corresponding to Species A-F can have an antigen expression gene. Thus, Species A and B are not mutually exclusive from any of Species C-G; Species C and D are not mutually exclusive from any of Species A, B, or E-G; Species E and F are not mutually exclusive from any of Species A-C or G; and Species G is not mutually exclusive from any of Species A-F. Species A and B, C and D, and E and F represent the only mutually exclusive species. From this, it can be seen that there can be no proper requirement involving election of Species G since there is no mutually exclusive alternative species encompassed by the claims. Applicants submit that, to the extent that the present species are the proper subject of an election requirement, the other, non-mutually exclusive "species" are encompassed by claims reciting a particular species. Accordingly, applicants submit that Species A, B, E, F, and G are encompassed by, and form a part of the examined subject matter, of claims reading on Species C. Only the subject matter of Species D should be withdrawn from consideration.

In regard to designation of generic claims, applicants refer to MPEP § 806.04(e) which states that "[c]claims may be restricted to a single disclosed embodiment (i.e. a single species, and thus be designated *a specific or species claim*), or a claim may include two or more of the disclosed embodiment...(and thus be designated *a generic or genus claim*)" (emphasis in original). Accordingly, applicants note that, with respect to species A and B, claims 1-5, 8-35, 37, and 38 are generic, with respect to species C and D, claims 1-13, 16-36, and 38 are generic, and with respect to species E and F, claims 1-17 and 20-37 are

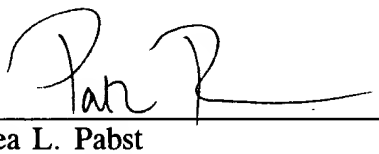
generic. With respect to species G, applicants note that claims 1-20, 23, 24, 27-30, and 33-38 are generic. In view of the discussion above, applicants assert that claims 1-14 and 16-38 read on the elected species and are subject to examination.

Applicants also traverse the restriction requirement as currently set forth for the following reasons. To be valid, a restriction requirement must establish both that (1) the "inventions" are either independent or distinct, and (2) that examination of more than one of the "inventions" would constitute a burden to the Examiner. The Office Action mailed October 18, 1996 sets forth reasons why the "species" are distinct (i.e. they are "structurally and functionally different"). Applicants note, however, that election of species should not be required if the species claimed would be considered unpatentable over each other (see MPEP § 808.01(a). Applicants urge that this point should be carefully considered by the Examiner in regard to the identified species. Notwithstanding this, applicants note that the restriction requirement does not provide sufficient basis to indicate that examination of more than one of the "species" would overly burden the Examiner. The only reason given is that a divergent literature and patent search would be required. With respect, applicant is unaware of any classes or subclasses specifying up or down regulation as a point of classification. In the art of gene regulation, generic regulation (up or down) is considered to be a single topic (with, of course, individual examples of various forms of regulation known). This can be seen by reference to nearly any textbook covering the topic of gene regulation. For example, Chapter 11 of Genes V (Lewin, John Wiley and Sons, New York, 1995), a copy of which is

attached, discusses gene regulation in general and illustrates gene regulation with a series of examples without dividing or identifying the different examples according to up or down regulation, nor with respect to the type of gene being regulated. Applicants also note that there is no discussion of, nor has it been established that, even if a divergent search were required, such a search would be overly burdensome *in this case*. For these reasons, applicants assert that election of the identified species is not proper.

Favorable consideration of claims 1-38 is earnestly solicited.

Respectfully submitted,



Patrea L. Pabst
Reg. No. 31,284

Date: November 18, 1996

ARNALL GOLDEN & GREGORY
2800 One Atlantic Center
1201 West Peachtree Street
Atlanta, Georgia 30309-3400

Certificate of Mailing under 37 CFR § 1.8(a)

I hereby certify that this Preliminary Amendment and Response to Restriction Requirement, along with any paper referred to as being attached or enclosed, is being deposited with the United States Postal Service on the date shown below with sufficient postage as first-class mail in an envelope addressed to the Assistant Commissioner for Patents, Washington, D.C. 20231.

Teresa Spratt

Date: November 18, 1996

Appendix

1. An isolated microbial cell comprising an Environmentally Limited Viability System, wherein the cell is viable in a permissive environment and non-viable in a non-permissive environment, the system comprising

(a) an essential gene, wherein expression of the gene in the cell is essential to the viability of the cell, the essential gene is expressed when the cell is in the permissive environment and is not expressed when the cell is in the non-permissive environment; and

(b) a lethal gene, wherein expression of the gene is lethal to the cell and the lethal gene is expressed when the cell is in the non-permissive environment but not when the cell is in the permissive environment.

2. The cell of claim 1 wherein the permissive environment comprises a temperature of about 37°C and the non-permissive environment comprises a temperature of less than about 30°C.

3. The cell of claim 1 wherein the permissive environment is inside a warm-blooded animal and the non-permissive environment is outside a warm-blooded animal.

4. The cell of claim 1 wherein the essential gene, the lethal gene, or both, is carried on an extrachromosomal vector.

5. The cell of claim 4 wherein the lethal gene is carried on an extrachromosomal vector and expression of the lethal gene is regulated by an expression product of a regulatory gene.

6. The cell of claim 5 wherein the expression product of the regulatory gene inhibits expression of the lethal gene and is expressed or active only in the permissive environment.

7. The cell of claim 5 wherein the expression product of the regulatory gene induces expression of the lethal gene and is expressed or active only in the non-permissive environment.

8. The cell of claim 4 wherein the vector has two lethal genes.

9. The cell of claim 8 wherein the vector comprises pMEG-104.

10. The cell of claim 1 wherein the cell is a gram-negative bacterium.

11. The cell of claim 10 wherein the gram-negative bacterium is an enteric bacterium.

12. The cell of claim 11 wherein the genus of the enteric bacterium is selected from the group consisting of *Escherichia* and *Salmonella*.

13. The cell of claim 1 wherein expression of the essential gene is regulated by an expression product of a regulatory gene.

14. The cell of claim 13 wherein the expression product of the regulatory gene inhibits expression of the essential gene and is expressed or active only in the non-permissive environment.

15. The cell of claim 13 wherein the expression product of the regulatory gene induces expression of the essential gene and is expressed or active only in the permissive environment.

16. The cell of claim 4 wherein the system further comprises a replication gene carried on a chromosome of the cell, the expression of which is required for replication of the vector, wherein the replication gene is expressed in the permissive environment and is not expressed in the non-permissive environment.

17. The cell of claim 16 wherein expression of the replication gene is regulated by an expression product of a regulatory gene.

18. The cell of claim 17 wherein the expression product of the regulatory gene inhibits expression of the replication gene and is expressed or active only in the non-permissive environment.

19. The cell of claim 17 wherein the expression product of the regulatory gene induces expression of the replication gene and is expressed or active only in the permissive environment.

20. The cell of claim 1 further comprising an expression gene wherein the expression gene encodes a desired expression product.

21. The cell of claim 20 wherein the desired expression product is an antigen.

22. The cell of claim 21 wherein the antigen is selected from the group consisting of bacterial antigens, viral antigens, plant antigens, fungal antigens, insect antigens, and non-insect animal antigens.

23. The cell of claim 1 for use as a vaccine, wherein the cell is viable when in the an animal and non-viable when outside of the animal, the essential gene is expressed when the cell is in the animal and is not expressed when the cell is outside of the animal, and the lethal gene is expressed when the cell is outside of the animal and is not expressed when the cell is in the animal, wherein the permissive environment comprises a temperature of about 37°C and the non-permissive environment comprises a temperature of less than about 30°C.

24. The cell of claim 23 further comprising an expression gene wherein the expression gene encodes a desired expression product.

25. The cell of claim 24 wherein the desired expression product is an antigen.

26. The cell of claim 25 wherein the antigen is selected from the group consisting of bacterial antigens, viral antigens, plant antigens, fungal antigens, insect antigens, and non-insect animal antigens.

27. A method of making a cell strain with environmentally limited viability comprising stably introducing into a cell

(a) an essential gene, wherein expression of the gene in the cell is essential to the viability of the cell, the essential gene is expressed when the cell is in the permissive environment and is not expressed when the cell is in the non-permissive environment;

(b) a lethal gene, wherein expression of the gene is lethal to the cell and the lethal gene is expressed when the cell is in the non-permissive environment but not when the cell is in the permissive environment,

wherein the cell strain is viable in a permissive environment and non-viable in a non-permissive environment.

28. The method of claim 27 wherein the permissive environment comprises a temperature of about 37°C and the non-permissive environment comprises a temperature of less than about 30°C.

29. The method of claim 27 wherein the permissive environment is inside a warm-blooded animal and the non-permissive environment is outside a warm-blooded animal.

30. A method of inducing immunoprotection in a warm-blooded animal comprising

administering to the animal a vaccine comprising a microbial cell comprising an Environmentally Limited Viability System, wherein the cell is viable when in the animal and non-viable when outside of the animal, the system comprising

(a) an essential gene, wherein expression of the gene in the cell is essential to the viability of the cell, the essential gene is expressed when the cell is in the animal and is not expressed when the cell is outside of the animal; and

(b) a lethal gene, wherein expression of the gene is lethal to the cell and the lethal gene is expressed when the cell is outside of the animal but not when the cell is in the animal.

31. The method of claim 30 wherein the system further comprising an expression gene wherein the expression gene encodes an antigen.

32. The method of claim 31 wherein the antigen is selected from the group consisting of bacterial antigens, viral antigens, plant antigens, fungal antigens, insect antigens, and non-insect animal antigens.

33. The method of claim 30 wherein the cell is administered to mucosal surfaces of the animal.

34. The method of claim 33 wherein the mucosal surfaces are in the gastrointestinal tract.

35. The method of claim 30 wherein the essential gene, the lethal gene, or both, is carried on an extrachromosomal vector, and wherein the system further comprises a replication gene carried on a chromosome of the cell, the expression of which is required for

replication of the vector, wherein the replication gene is expressed when the cell is in the animal and is not expressed when the cell is outside of the animal.

36. The cell of claim 5 wherein the absence of a functional expression product of the regulatory gene derepresses expression of the lethal gene and wherein the expression product is not expressed or is inactive only in the non-permissive environment.

37. The cell of claim 13 wherein the absence of a functional expression product of the regulatory gene derepresses expression of the essential gene and wherein the expression product is not expressed or is inactive only in the permissive environment.

38. The cell of claim 17 wherein the absence of a functional expression product of the regulatory gene derepresses expression of the replication gene and wherein the expression product is not expressed or is inactive only in the permissive environment.